

In the Claims:

Please cancel claims 1-13.

Please amend claims 14 and 15 as follows:

14. (currently amended) ~~The~~ An imaging apparatus, comprising of  
~~claim 18, wherein the processor further:~~

a motorized stage;

a camera having a lens directed toward said motorized stage; and

a processor coupled to said camera, wherein said processor contains  
instructions which, when executed by said processor, cause said processor to:

select at least three points of a sample adjacent said motorized  
stage;

determine a stage position for each selected point;

focus said camera on each selected point;

determine an object distance from the camera lens to the sample at  
each selected point;

develop a focus surface based on stage position and object  
distance for the at least three selected points;

determines for a selected point whether an error function based on  
the difference between the object distance at the selected point and the object  
distance of the surface at that point is greater than a predetermined limit;

deletes the point from the selection if the error is greater than the  
predetermined limit; and

develops a focus surface based on stage position and object  
distance for the remaining selected points.

15. (currently amended) ~~The~~ An imaging apparatus, comprising of  
~~claim 18, wherein the processor further:~~

a motorized stage;

a camera having a lens directed toward said motorized stage; and

a processor coupled to said camera, wherein said processor contains  
instructions which, when executed by said processor, cause said processor to:

select at least three points of a sample adjacent said motorized  
stage;

determine stage position for each selected point;

focus said camera on each selected point;

determine object distance from the camera lens to the sample at  
each selected point;

develop a focus surface based on stage position and object  
distance for the at least three selected points;

determines for a selected point whether an error function based on  
the difference between the object distance at the selected point and the object  
distance of the surface at that point is greater than a predetermined limit;

selects an additional point from an area in which the point has an  
error function greater than a predetermined limit;

determines stage position for the additional selected point;

focuses said camera on the additional selected point;

determines object distance from the camera lens to the sample at  
the additional selected point; and

develops a focus surface based on stage position and object  
distance for the selected points.

Please cancel claims 16-18.

19. (previously presented) A system for creating a high throughput montage image of a microscope slide, the system comprising:

an optical system that comprises at least one camera, a motorized stage for moving a slide while an image of the slide is captured, a pulsed light illumination system that optically stops motion on the motorized stage while allowing continuous physical movement of the motorized stage, and a stage position detector that controls firing of the pulsed light illumination system at predetermined positions of the motorized stage;

a first component that identifies sample regions on the slide in the optical system and determines locations of the sample on the slide, wherein the first component uses information about the locations to generate control parameters for the motorized stage and the camera;

a second component that uses the control parameters to ensure that a high-quality montage image is captured; and

means for capturing a montage image, using the second component, by maintaining motion of the motorized stage and synchronization of the optical system, thereby enabling accurate focus control of optical elements without requiring the stage to be stopped and refocused at each tile location.

20. (original) The system of claim 19, wherein the optical system is a bright field microscope.

21. (previously presented) The system of claim 19, wherein the pulsed light illumination system is a standard strobe light, wherein stage location is determined by the stage position detector, wherein the stage location executes the strobe light and wherein the system does not depend on uniform motion of the motorized stage over an imaged area to execute the strobe light.

22. (original) The system of claim 21, wherein the camera is free running and the motorized stage speed is matched to the camera's frame rate to an extent that prevents execution of the pulse light illumination system from falling outside of an exposure window.

23. (original) The system of claim 19, wherein the pulse light illumination system is any pulsed light source.

24. (original) The system of claim 19, wherein the stage position detector is a Ronchi ruler that is attached to the motorized stage, wherein the Ronchi ruler is a pattern of alternating light and dark bands that are equally spaced along a substrate.

25. (original) The system of claim 24, wherein the stage position detector utilizes a light sensor that is mechanically isolated from the Ronchi ruler, wherein as the ruler passes under the sensor, a series of electronic pulses that correspond to the alternating light and dark bands of the Ronchi ruler is generated and the series of electronic pulses is used to monitor the position and direction of the motorized stage.

26. (original) The system of claim 19, wherein the position system uses only position information from a stage controller without an external feedback sensor.

27. (previously presented) The system of claim 19 wherein the pulse illumination system is fired whenever the stage position detector determines that the motorized stage has moved into a neighboring field of view of the camera.

28. (original) The system of claim 19, wherein signals from the stage position detector represent motions of the motorized stage, and wherein timing of the signals vary depending on speeds of the motorized stage.

29. (original) The system of claim 19, wherein an absolute speed of the motorized stage is not relevant.

30. (original) The system of claim 21, wherein a stage position as determined by the stage position detector executes the camera and the pulse light illumination system, wherein the camera is not free running and each camera frame corresponds to an equally spaced positional change that is independent of a stage velocity.

31. (previously presented) The system of claim 19, wherein the first component includes an image cropping component for identifying sample regions on the slide to be scanned, wherein the image cropping component:  
determines a location of a slide boundary by searching intervals corresponding to regions expected to contain edges of the slide; and  
removes portions of the image falling outside of the determined slide boundary.

32. (previously presented) The system of claim 31, wherein the image cropping component converts a copy of the image to a grayscale image.

33. (previously presented) The system of claim 31, wherein the image cropping component identifies pixel blocks that are likely to contain remaining boundary edges and flag the blocks as edges that should not be considered for high-resolution imaging.

34. (previously presented) The system of claim 19, wherein the first component includes a sample finding component that locates regions in the image that contain the sample of interest.

35. (previously presented) The system of claim 34, wherein a cropped image is inputted into the sample finding component from an image cropping component, wherein the sample finding component identifies sample regions by applying a sequence of filters that incorporate knowledge of typical appearance and location of sample and non-sample slide regions and outputs a matrix having values that indicate which regions should be imaged.

36. (previously presented) The system of claim 19, wherein a filter analyzes mean pixel intensity to generate a threshold value for making an initial classification of sample versus non-sample regions.

37. (previously presented) The system of claim 19, wherein a filter analyzes a difference between pixel intensities to generate a threshold value for making a classification of sample versus non-sample regions.

38. (previously presented) The system of claim 19, wherein a filter analyzes mean and standard deviation of local pixel intensities and combines the mean and the standard deviation to generate a threshold value for making a classification of sample versus non-sample regions.

39. (previously presented) The system of claim 19, wherein the intensities are used to differentiate sample-containing regions from blank regions and other non-sample containing regions and the standard deviation represents the amount of variation in pixel values and is therefore an indicator of the border between the sample and the blank slide.

40. (previously presented) The system of claim 19, wherein morphological filters are applied to an array representing selected and unselected regions which in turn represent sample and non-sample regions to refine classification based on size and position of neighboring groups of potential sample pixels, wherein the morphological filters process pixels of the image in groups that correspond to slide regions that can be imaged individually during a high-resolution scanning process.

41. (previously presented) The system of claim 40, wherein the morphological filters ensure that tiles that are partially filled with the sample are classified as sample-containing tiles.

Please cancel claim 42.

43. (previously presented) The system of claim 19, wherein the first component includes a scan control component that interprets a matrix, outputted by a sample finding component, and transposes positions of the matrix into actual stage coordinates for a microscopic imaging.

44. (previously presented) The system of claim 19, wherein the second component comprises:

a focus point selection component that evaluates sample regions of an image and selects several points on which to initially focus microscope optics on a point-by-point basis;

a focal surface determination component that uses focus positions to generate a three-dimensional data set corresponding to optical specimen distance at each stage location, wherein data points in the data set are input to a routine that generates control parameters for a slide scanning process; and

a scan component that captures the montage image by maintaining motion of a stage and synchronization of a microscopic imaging system during montage image acquisition, thereby enabling accurate focus control of optical elements without requiring stopping and refocusing of the stage at each tile location and substantially reducing montage acquisition time.

45. (previously presented) The system of claim 44, wherein the focus point selection component selects positions based on their relative contrast within the image and their overall distribution with respect to a sample coverage area.

46. (original) The system of claim 44, wherein focus points are definable through an input file.

47. (original) The system of claim 44, wherein focus points are definable through a user interface.

48. (previously presented) The system of claim 44, wherein focus points are predefined and repeated for each slide without any preprocessing to find sample regions, when sample locations are reproducible on the slides.

49. (original) The system of claim 44, wherein the number of data points required depends on the actual three-dimensional structure defined by the specimen and the slide and the geometrical dimension of the surface to be fit.

50. (previously presented) The system of claim 49, wherein once the surface is determined, an error function is calculated to determine a fit accuracy, and wherein if the accuracy indicated by the error function exceeds expected limits, additional points can be acquired and the surface recalculated.

51. (original) The system of claim 44, wherein the specimen is positioned such that each camera image is aligned within the equivalent single pixel spacing.

52. (original) The system of claim 44, wherein to maintain focus during the scanning process, the stage is positioned at a proper focal position as determined by the focus surface parameters.

53. (previously presented) A method for creating a high throughput montage image of microscope slides, the method comprising:

placing a slide to be imaged in a slide holder on a motorized stage;

capturing a low resolution image of the slide;

identifying sample locations within the image;

generating control parameters to scan regions of interest under microscopic optics;

capturing a high resolution montage image by enabling accurate focus control of optical elements without requiring that the motorized stage be stopped and refocused at each tile location in the montage image;

controlling a tiling process by moving the motorized stage;



capturing image tiles with precise alignment by executing a strobe illumination system whenever a stage position sensor determines that the motorized stage has moved to a neighboring field of view of a camera;  
scanning each row of locations identified to contain the sample for the sample; and  
removing the slide and inserting another slide to be imaged.

54. (previously presented) The method of claim 53, wherein identifying further comprises:

flat field correcting the image using a blank slide and a similar image obtained from a camera that captured the image;  
cropping the image by an image cropping component;  
inputting a cropped image into a sample finding component, wherein the sample finding component identifies sample regions by applying a sequence of filters that incorporate knowledge of typical appearance and location of sample and non-sample slide regions and outputs a tiling matrix whose values indicate which tiles should be imaged; and  
interpreting the tiling matrix, by a scan control component, and transposing positions of the tiling matrix into an actual stage coordinate for a microscopic imaging.

55. (previously presented) The method of claim 53, wherein generating further comprises:

evaluating sample regions of the low resolution image and selecting several focus positions on which to initially focus microscope optics on a point-by-point bases;  
placing under the microscope optics each focus position, wherein the best-fit focus at each position is determined;  
generating a three-dimensional data set corresponding to an optimal sample distance at each stage location, wherein data points in the data set are input to a routine that generates necessary control parameters for a slide scanning process; and

passing the sample information and control parameters to a component that is responsible for the motion of a stage and synchronization of a microscopic imaging system during a montage image acquisition.

56. (previously presented) The method of claim 55, further comprising determining a surface on the slide that represents a focal position of the sample and using information from the surface to automatically control focus as the sample is scanned under microscope optics.

57. (previously presented) The method of claim 55, wherein evaluating further comprises selecting positions based on their relative contrast within the low resolution image and their overall distribution with respect to a sample coverage area.

58. (original) The method of claim 55, wherein evaluating further comprises allowing a user to define focus points.

59. (previously presented) The method of claim 55, wherein generating the number of data points required depends on the actual three-dimensional structure defined by the sample and the slide and the geometrical dimension of the surface to be fit, wherein an error function is calculated to determine a fit accuracy once the surface has been determined, and if the accuracy exceeds expected limits additional points can be acquired and the surface recalculated.

60. (previously presented) The method of claim 55, further comprising positioning the sample such that each camera image is aligned with the equivalent single pixel spacing.

61. (original) The method of claim 55, further comprising positioning the stage at a proper focal position as determined by focus surface parameters.

62. (original) The method of claim 55, further comprising computing a vertical velocity, as a function of a parameter and subsequently of time, from the derivative of the focal surface if imaging is accomplished during continuous motion of the stage in a x-direction via an imaging arrangement.

63. (original) The method of claim 62, further comprising using the velocity to control the optical position and maintain focus as images are acquired continuously across a row.

64. (original) The method of claim 62, further comprising requiring a new velocity function for each row scanned based on a stepped y-position.

65. (previously presented) The method of claim 54, wherein cropping further comprises the steps of:

determining an approximate location of a slide boundary by searching intervals corresponding to regions expected to contain edges of the slide; and removing portions of the image falling outside of the determined slide boundary.

66. (previously presented) The method of claim 65, further comprising converting a copy of the low resolution image to a grayscale image.

67. (previously presented) The method of claim 65, further comprising identifying pixel blocks that are likely to contain remaining boundary edges and flagging these blocks as edges that should not be considered for high resolution imaging.

68. (original) The method of claim 65, further comprising analyzing mean and standard deviation of local pixel intensities and combining the mean and the standard deviation to generate a threshold value.

69. (previously presented) The method of claim 68, further comprising using the intensities to differentiate sample-containing regions from blank regions and other non-sample containing regions.

70. (previously presented) The method of claim 65, further comprising applying morphological filters to threshold standard deviation data to refine classification based on size and position of neighboring groups of potential sample pixels, whereby the morphological filters process pixels of the cropped image in groups that correspond to slide regions that can be imaged individually during a high-resolution scanning process.

71. (original) The method of claim 53, wherein controlling comprises attaching a Ronchi ruler to the motorized stage.

72. (original) The method of claim 71, wherein controlling further comprises utilizing a light sensor that is mechanically isolated from the Ronchi ruler, whereby as the Ronchi ruler passes under the light sensor a series of electronic pulses that corresponds to alternating light and dark bands of the Ronchi ruler is generated.

73. (original) The method of claim 53, wherein capturing further comprises capturing images of tiles with precise alignment until a row is finished.

74. (original) The method of claim 53, wherein capturing further comprises capturing images of tiles with precise alignment until a controlling program tells the system to stop.

Please add new claim 75 as follows:

75. (new) The imaging apparatus of claim 15, wherein the processor further deselects the point having an error function greater than the predetermined limit.